

Understanding the impacts of anthropogenic sound on beaked whales¹

T.M. COX^{*†}, T.J. RAGEN^{*}, A.J. READ⁺, E. VOS^{*}, R.W. BAIRD[#], K. BALCOMB^{**}, J. BARLOW⁺⁺, J. CALDWELL^{##}, T. CRANFORD[^], L. CRUM^{^^}, A. D'AMICO[✉], G. D'SPAIN[§], A. FERNÁNDEZ[¥], J. FINNERAN[✉], R. GENTRY^{§§}, W. GERTH^{✉✉}, F. GULLAND[§], J. HILDEBRAND[§], D. HOUSER[□], T. HULLAR[±], P.D. JEPSON[□], D. KETTEN[•], C.D. MACLEOD[♀], P. MILLER[□], S. MOORE[✉], D.C. MOUNTAIN[◇], D. PALKA[▲], P. PONGANIS[§], S. ROMMEL[■], T. ROWLES[◆], B. TAYLOR⁺⁺, P. TYACK[•], D. WARTZOK^{○○}, R. GISINER^{••}, J. MEAD^Σ, L. BENNER^{□□}

Contact e-mail: tara.cox@duke.edu

This paper is dedicated to the memory of Dr. Edward Thalmann (1945-2004).

ABSTRACT

This review considers the effect of anthropogenic sound on beaked whales². Two major conclusions are presented: (1) gas-bubble disease, induced in supersaturated tissue by a behavioural response to acoustic exposure, is a plausible pathologic mechanism for the morbidity and mortality seen in cetaceans associated with sonar exposure and merits further investigation; and (2) current monitoring and mitigation methods for beaked whales are ineffective for detecting these animals and protecting them from adverse sound exposure. In addition, four major research priorities, needed to address information gaps on the impacts of sound on beaked whales, are identified: (1) controlled exposure experiments to assess beaked whale responses to known sound stimuli; (2) investigation of physiology, anatomy, pathobiology and behaviour of beaked whales; (3) assessment of baseline diving behaviour and physiology of beaked whales; and (4) a retrospective review of beaked whale strandings.

KEYWORDS: BEAKED WHALES; ZIPHIIDAE; NOISE; MANAGEMENT; ACOUSTICS; CONSERVATION; STRANDINGS

¹ This paper arose out of a workshop convened in April 2004 by the US Marine Mammal Commission in Baltimore, Maryland, USA.

² Diverse fields (marine mammal ecology, medicine, behaviour, physiology, pathobiology and anatomy, human diving physiology and acoustics) were represented at the 2004 workshop and were brought together for an interdisciplinary discussion of various topics related to interactions between beaked whales and anthropogenic sound. These included sound propagation and acoustic exposure during specific stranding events, behaviour and ecology of beaked whales, beaked whale distribution, abundance and habitat, beaked whale anatomy and physiology and the efficacy of existing monitoring and mitigation efforts.

INTRODUCTION

Beaked whales (family Ziphiidae) are among the least understood marine mammals. The family consists of approximately 21 species that spend relatively little time at the surface and occur almost exclusively in deep waters beyond the continental shelf. Most of our current knowledge of beaked whales is based on studies of stranded specimens. Reports of occasional mass strandings of beaked whales (i.e. strandings of two or more whales other than a cow-calf pair,

* US Marine Mammal Commission, 4340 East-West Highway, Rm. 905, Bethesda, MD 20814, USA.

† Current address: Duke University Marine Laboratory, 135 Duke Marine Lab Road, Beaufort, NC 28516, USA.

+ Duke University Marine Laboratory, 135 Duke Marine Lab Road, Beaufort, NC 28516, USA.

Cascadia Research Collective, 218 12W. 4th Avenue, Olympia, WA 98501, USA.

** Center for Whale Research, Inc., 355 Smugglers Cove Road, Friday Harbor, WA 98250, USA.

++ Southwest Fisheries Science Center, National Marine Fisheries Service, 8604 La Jolla Shores Dr., La Jolla, CA 92037-1508, USA.

Schlumberger Reservoir Service, Data and Consulting Services, 3600 Briarpark Drive, Houston, TX 77042-5206, USA.

^ San Diego State University, Biology Department, 5500 Campanile Drive, San Diego, CA 92182-0001, USA.

^^ Center for Industrial and Medical Ultrasound, Applied Physics Laboratory, University of Washington, 1013 NE 40th Street, Seattle, WA 98105, USA.

✉ SPAWAR Systems Center – San Diego, 53560 Hull Street, San Diego, CA 92152-5001, USA.

§ Scripps Institution of Oceanography, 291 Rosencrans St., San Diego, CA 92106, USA.

¥ Institute of Animal Health, Veterinary School, Departamento de Morfología, Edificio de Veterinaria, Universidad de Las Palmas de Gran Canaria, Spain.

§§ Office of Protected Resources, National Marine Fisheries Service, 1315 East-West Highway, Silver Spring, MD 20910, USA.

✉✉ Navy Experimental Diving Unit, 321 Bullfinch Road, Panama City, FL 32407-7015, USA.

□ The Marine Mammal Center, 1065 Fort Cronkhite, Sausalito, CA 94965, USA.

□□ Biomimetica, 7951 Shantung Drive, Santee, CA 92071, USA.

± Washington University School of Medicine, Department of Otolaryngology, 660 South Euclid Avenue, Box 8115, St. Louis, MO 63110, USA.

□ Institute of Zoology, Zoological Society of London, Regents Park, London NW1 4RY, UK.

● Woods Hole Oceanographic Institution, Biology Department, MS #50, Woods Hole, MA 02543, USA.

♀ University of Aberdeen, School of Biological Sciences, Department of Zoology, Tillydrone Avenue, Aberdeen, AB24 2TZ, UK.

□ Sea Mammal Research Unit, Gatty Marine Laboratory, University of St. Andrews, St. Andrews, Fife, KY16 8LB, Scotland, UK.

✉ Alaska Fisheries Science Center, 7600 Sand Point Way NE, Seattle, WA 98115-0070, USA.

◇ Boston University, Department of Biomedical Engineering, 44 Cummington Street, Boston, MA 02215, USA.

▲ Northeast Fisheries Science Center, National Marine Fisheries Service, 166 Water Street, Woods Hole, MA 02543-1026, USA.

■ Florida Fish and Wildlife Conservation Commission, Florida Wildlife Research Institute, Marine Mammal Pathology Lab., 3750 54th Avenue South, St. Petersburg, FL 33711, USA.

◆ Hollings Marine Laboratory, 331 Ft. Johnson Road, Charleston, SC 29412, USA.

○○ Florida International University, University Park, Miami, FL 33199, USA.

•• Office of Naval Research, 875 Randolph St., Suite 1425, Alexandria, VA 22217-5660, USA.

Σ National Museum of Natural History, 10th and Constitution Avenue, NW, Washington DC 20560, USA.

□□ US Commission on Ocean Policy, 1120 20th Street, NW, Washington DC 20036, USA.

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Geraci and Lounsbury, 1993) date back to at least the early 1800s. Since 1960, however, 41 'mass' strandings of Cuvier's beaked whales (*Ziphius cavirostris*) have been reported worldwide (Brownell *et al.*, 2004; Taylor *et al.*, 2004). Furthermore, these probably represent only a small proportion of all beaked whale strandings. Some of these recent mass strandings were concurrent with naval manoeuvres and the use of active sonar (Frantzis, 1998³; Anon., 2001; Jepson *et al.*, 2003). The overall pattern of strandings has raised concerns that certain sounds from sonar could directly or indirectly result in the death or injury of beaked whales, particularly Cuvier's beaked whales. Additional concerns have been raised that sounds from seismic surveys might have similar effects (Taylor *et al.*, 2004).

Recent stranding events

Several recent mass strandings have led to suggestions that exposure to anthropogenic sounds negatively affects beaked whales. The temporal and spatial association of mass strandings of beaked whales and offshore naval manoeuvres was first noted in 1991 (Simmonds and Lopez-Jurado, 1991). Since then, a series of 'atypical' (Frantzis, 1998) beaked whale strandings, temporally (within hours or days) and spatially (less than 50km) associated with naval manoeuvres, have been better documented and are briefly summarised below. These strandings lend further support to the hypothesis that exposure to certain anthropogenic sounds may harm these animals.

Greece, May 1996

Frantzis (1998) reported an 'atypical' mass stranding of 12 Cuvier's beaked whales on the coast of Greece that was associated with acoustic trials by vessels from the North Atlantic Treaty Organisation (NATO). He was the first to hypothesise that these strandings were related to exposure to low-frequency military sonar. However, the sonar in question produced both low- and mid-frequency signals (600Hz, 228dB Sound Pressure Level (SPL) (re: 1µPa at 1m Root Mean Square (RMS))⁴, 3kHz, 226dB SPL, D'Amico and Verboom, 1998). Frantzis's hypothesis prompted an in-depth analysis of the acoustic activity during the naval exercises, the nature of the strandings and the possibility that the acoustic source was related to the strandings (D'Amico and Verboom, 1998). Since full necropsies had not been conducted and no gross or histological abnormalities were noted, the cause of the strandings could not be determined unequivocally (D'Amico and Verboom, 1998). The analyses thus provided some support but no clear evidence for the hypothesised cause-and-effect relationship of sonar operations and strandings.

Bahamas, March 2000

When multiple beaked whales atypically stranded in the Bahamas in March 2000, researchers were aware of the possible link to anthropogenic sound sources and thus facilitated a more comprehensive examination of the dead animals. However, in most cases, analyses were performed on decomposed carcasses or tissues. Seventeen cetaceans (one spotted dolphin, *Stenella frontalis*, nine Cuvier's beaked whales, three Blainville's beaked whales,

Mesoplodon densirostris, two minke whales, *Balaenoptera acutorostrata* and two unidentified beaked whales) stranded on 15-16 March 2000 on beaches of the Bahamas Islands. Eight beaked whales were returned to the water alive and one dead specimen was not readily accessible for necropsy. As a result, only five of the stranded beaked whales were examined *post mortem* and only two of these were marginally fresh enough to allow a more detailed pathological analysis of lesions. Initial gross necropsy of these five beaked whales indicated that the animals were in good body condition and that none presented any gross indication of debilitating infectious disease. Computerised tomography of two animals and detailed dissection of five heads indicated subarachnoid haemorrhages in the temporal region and haemorrhage in the cochlear duct of two of the animals. The *post mortem* time to examination varied from hours to several days, unfortunately compromising these analyses. The interim report of the investigation concluded that these findings were consistent with acoustic or impulse injuries that resulted in the animals stranding. The gross and histopathological evidence indicated cardiovascular collapse, which is often associated with other signs of extreme physiological stress observed in live, beach-stranded marine mammals (i.e. hyperthermia, high endogenous catecholamine release; Anon., 2001; see also Balcomb and Claridge, 2001). The role of intracranial and acoustic fat injuries in the strandings and mortalities was not clear. Analysis of acoustic sources used in the Bahamas naval exercises revealed that four of five ships were using mid-frequency sonar (AN/SQS-53C: 2.6-3.3kHz, ~235dB SPL, AN/SQS-56: 6.8, 7.5 and 8.2kHz, ~223dB SPL; Anon., 2001). The final report of the joint US National Oceanographic and Atmospheric Administration (NOAA) and US Navy investigation into the stranding event, including the full suite of pathological investigations is still pending. The event raised the question of whether the mid-frequency component of the sonar in Greece in 1996 was implicated in the stranding, rather than the low-frequency component proposed by Frantzis (1998).

Madeira, May 2000

The stranding in the Bahamas was soon followed by another atypical mass stranding of Cuvier's beaked whales in the Madeira Islands. Between 10 and 14 May 2000, three Cuvier's beaked whales stranded on two islands in the Madeira archipelago. NATO naval exercises involving multiple ships occurred concurrently with these strandings, although NATO has thus far been unwilling to provide information on the sonar activity during their exercises. Only one of the stranded animals was marginally fresh enough for a full necropsy (24 hours post-stranding). The necropsy revealed evidence of haemorrhage and congestion in the right lung and both kidneys (Freitas, 2004), as well as evidence of intracochlear and intracranial haemorrhage similar to that observed in the Bahamas beaked whales (D. Ketten, unpublished data).

Canary Islands, September 2002

In September 2002, a beaked whale stranding event occurred in the Canary Islands. On 24 September, 14 beaked whales (7 Cuvier's beaked whales, 3 Blainville's beaked whales, 1 Gervais' beaked whale, *M. europaeus*, and 3 unidentified beaked whales) stranded on the beaches of Fuerteventura and Lanzarote Islands, close to the site of an international naval exercise (called Neo-Tapon 2002) held that same day. The first strandings began about four hours after the onset of the use of mid-frequency sonar activity (3-

³ Frantzis (1998) cited the following references: Robinson *et al.* (1983); Miyazaki (1989); Simmonds and Lopez-Jurado (1991) and Tortonesi (1963).

⁴ Unless otherwise noted, all SPL values are RMS pressures referenced to 1µPa at 1m.

10kHz, D'Spain *et al.*, 2006; Jepson *et al.*, 2003). Seven whales (1 female Blainville's beaked whale, 1 female Gervais' beaked whale and 5 male Cuvier's beaked whales) are known to have died that day (Fernández *et al.*, 2005). The remaining seven live whales were returned to deeper waters. Over the next three days, three male and one female Cuvier's beaked whales were found dead and a carcass of an unidentified beaked whale was seen floating offshore. A total of nine Cuvier's beaked whales, one Blainville's beaked whale and one Gervais' beaked whale were examined post mortem and studied histopathologically (one Cuvier's beaked whale carcass was lost to the tide). No inflammatory or neoplastic processes were noted grossly or histologically and no pathogens (e.g. protozoa, bacteria and viruses, including morbillivirus) were identified. Stomach contents were examined in seven animals and six of them had recently eaten, possibly indicating that the event(s) leading to their deaths had had a relatively sudden onset (Fernández *et al.*, 2005). Macroscopic examination revealed that the whales had severe, diffuse congestion and haemorrhages, especially in the fat in the jaw, around the ears, in the brain (e.g. multifocal subarachnoid haemorrhages) and in the kidneys (Fernandez, 2004; Fernandez *et al.*, 2004). Gas bubble-associated lesions were observed in the vessels and parenchyma (white matter) of the brain, lungs, subcapsular kidney veins and liver; fat emboli were observed in epidural veins, liver sinusoids, lymph nodes and lungs (Jepson *et al.*, 2003; Fernandez, 2004; Fernandez *et al.*, 2004; 2005). After the event, researchers from the Canary Islands examined past stranding records and found reports of eight other strandings of beaked whales in the Canaries since 1985, at least five of which coincided with naval activities offshore (Martín *et al.*, 2004).

Gulf of California, September 2002

In September 2002, marine mammal researchers vacationing in the Gulf of California, Mexico discovered two recently deceased Cuvier's beaked whales on an uninhabited island. They were not equipped to conduct necropsies and in an attempt to contact local researchers, found that a research vessel had been conducting seismic surveys approximately 22km offshore at the time that the strandings occurred (Taylor *et al.*, 2004). The survey vessel was using three acoustic sources: (1) seismic air guns (5–500Hz, 259dB re: 1μPa Peak to Peak (p-p); Federal Register, 2003); (2) sub-bottom profiler (3.5kHz, 200dB SPL; Federal Register, 2004); and (3) multi-beam sonar (15.5kHz, 237dB SPL; Federal Register, 2003). Whether or not this survey caused the beaked whales to strand has been a matter of debate because of the small number of animals involved and a lack of knowledge regarding the temporal and spatial correlation between the animals and the sound source. This stranding underlines the uncertainty regarding which sound sources or combinations of sound sources may cause beaked whales to strand.

Although some of these stranding events have been reviewed in government reports or conference proceedings (e.g. Anon., 2001; Evans and Miller, 2004), many questions remain. Specifically, the mechanisms by which beaked whales are affected by sound remain unknown. A better understanding of these mechanisms will facilitate management and mitigation of sound effects on beaked whales. As a result, in April 2004, the United States Marine Mammal Commission (MMC) convened a workshop of thirty-one scientists from a diverse range of relevant disciplines (e.g. human diving physiology and medicine, marine mammal ecology, marine mammal anatomy and

physiology, veterinary medicine and acoustics) to explore issues related to the vulnerability of beaked whales to anthropogenic sound. The purpose of the workshop was to (1) assess the current knowledge of beaked whale biology and ecology and recent beaked whale mass stranding events; (2) identify and characterise factors that may have caused the strandings; (3) identify ways to more adequately investigate possible cause and effect relationships; and (4) review the efficacy of existing monitoring and mitigation methods. This paper arose out of the discussions at that workshop.

OVERVIEW OF RECENT FINDINGS

A number of scientists have prepared papers describing acoustic activities and propagation characteristics during some stranding events, beaked whale biology including behaviour and ecology, distribution, abundance, anatomy and physiology and mitigation and management (Barlow and Gisinier, 2006; Barlow *et al.*, 2006; D'Spain *et al.*, 2006; Ferguson *et al.*, 2006; MacLeod and D'Amico, 2006; MacLeod *et al.*, 2006 and Rommel *et al.*, 2006). These are briefly summarised here and where appropriate, we have made some general recommendations for further research topics.

Acoustic characteristics

D'Spain *et al.* (2006) described the acoustic sources and propagation parameters associated with several of the stranding events: Greece, 1996, the Bahamas, 2000 and the Canary Islands, 2002. The authors found that these three events shared three common features. One common environmental feature was deep water close to land (e.g. offshore canyons). Whether this feature influenced beaked whale distribution (e.g. species that stranded prefer this habitat), accentuated the effects of the sounds through reflection and reverberation from the bathymetry and/or acted in some other way is not clear. A second environmental feature common to all three events was the presence of an acoustic waveguide (see D'Spain *et al.*, 2006 for more details). Thirdly, the authors noted common transmission characteristics, including periodic sequences of transient pulses (i.e. rapid onset and decay times) generated at depths shallower than 10m (in the Bahamas and Canaries) by sound sources moving at speeds of 2.6m s⁻¹ or more during source operation (see table 1 in D'Spain *et al.*, 2006 for more details).

The sound sources in use during the Gulf of California stranding event in September 2002 included both a sub-bottom profiler and multi-beam sonar system (Table 1). Air guns can neither be confirmed nor ruled out as a cause of these strandings and retrospective analyses are needed to investigate the possible role of these other sound sources in the stranding event.

It is not yet clear whether high-intensity sound sources alone are sufficient to trigger beaked whale strandings, or whether certain acoustic, biological or environmental characteristics must co-occur with these stimuli. A more complete understanding of the source characteristics and propagation of anthropogenic sounds associated with beaked whale strandings would be extremely useful in predicting and preventing future incidents of this nature, but it is often difficult to obtain such specific information, which may be sensitive for many of the parties involved.

Based on the available data, we recommend research to: (1) identify key characteristics of sound (e.g. frequency, amplitude, energy, directional transmission pattern, use of

arrays vs. single sources, etc.) that may affect beaked whales; (2) identify characteristics of anthropogenic sounds associated with historic stranding events; (3) estimate the possible range of sound levels the animals received prior to stranding; (4) characterise environmental parameters that influence sound propagation and model site-specific sound propagation (on *post hoc* and predictive basis), especially where detailed environmental data are not immediately available; and (5) measure the behavioural responses of beaked whales in the presence of sound.

Behaviour and ecology

MacLeod and D'Amico (2006) reviewed the behaviour and ecology of beaked whales and their relevance to the impacts of sound on beaked whales. Specifically, they reviewed beaked whale social structure, life history, ecology, sound production and function and the characteristics of their habitat. Multiple strandings of beaked whales that occur concurrently with sound-generating anthropogenic activities often include a large proportion of immature and juvenile animals. However, it is not known whether juveniles are disproportionately affected, the age structure observed in the strandings is representative of that in beaked whale populations, or the strandings indicate geographic separation of demographic groups. If juveniles are disproportionately affected, it might suggest a relationship between the dimensions of some part of the anatomy and the wavelength of the sound involved or, alternatively, an age-specific behavioural response.

Recent tagging data from Cuvier's and Blainville's beaked whales from the Mediterranean Sea and Canary Islands (Tyack, unpub. data) have revealed several notable features of their dive profiles: (1) dives to depths near 2km and lasting nearly 1.5hrs; (2) slow ascent rates; and (3) a series of 'bounce' dives to 100-400m between the deeper, longer dives. The implications of this dive pattern are discussed below. We recommend a combination of short-(hours to days) and long-term studies (weeks to months) on the behaviour of beaked whales using multiple methods (e.g. D-tags which measure received sound levels as well as other movement data, time-depth recorders and visual observations) to better describe 'normal' behaviour.

Distribution, abundance and habitat

Barlow *et al.* (2006), Ferguson *et al.* (2006) and MacLeod *et al.* (2006) reviewed global distributions and abundance of beaked whales. Our understanding of the distribution of many beaked whales is very limited and based primarily on observations of strandings and a limited number of at-sea sightings. The identification of important habitat is generally compromised by insufficient and inconsistent observation effort. It is clear that research effort must focus on: (1) population structure, possibly using genetic data from archived samples (bone, skin, etc.) housed in museums and other collections around the world; and (2) population distributions.

Estimates of abundance and density are hindered by the typical surfacing behaviour of beaked whales at sea: their blows are generally not visible, they have low surfacing profiles and they spend the majority of their time at depth (Hooker and Baird, 1999; Baird *et al.*, 2004; Barlow and Gisinier, 2006). In addition, beaked whales tagged by Johnson *et al.* (2004) vocalised only when they were deeper than 200m, an observation that has important implications for passive acoustic monitoring. The importance of identifying, classifying and understanding vocalisations of beaked whales and the potential utility of passive acoustic monitoring must be noted. For such monitoring to be effective, future research must (1) develop and test detection algorithms; (2) ground-truth detection methods by coupling visual and passive acoustic studies and by monitoring vocalisations in areas for which there are good density estimates; and (3) investigate the behavioural context of vocalisations. In addition, it is important that effort be expended on: (1) estimation of abundance and densities of beaked whale species, especially in those areas where sound-producing activities are planned or regularly carried out; (2) systematic surveys that include oceanographic data to help identify key habitat characteristics; and (3) increase understanding of movement patterns via multiple methods (e.g. telemetry).

An improved understanding of basic beaked whale biology will advance the potential for predictive habitat modelling and may help managers and sound-producers predict which areas support high densities of beaked whales

Table 1
Acoustic characteristics of sound sources and propagation during stranding events.

Name	Centre frequency	SPL	Beam direction	Model propagation?
Greece¹, May 1996				
TVDS	600Hz	228dB re: 1µPa at 1m (RMS)	Horizontal	Yes
TVDS	3kHz	226dB re: 1µPa at 1m (RMS)	Horizontal	Yes
Bahamas², March 2000				
AN/SQS 53C	2.6kHz; 3.3kHz	~235dB re: 1µPa at 1m (RMS)	3° down from horizontal	Yes
AN/SQS 56	6.8kHz; 7.5kHz; 8.2kHz	223dB re: 1µPa at 1m (RMS)	Horizontal	Yes
Madeira, May 2000				
Unknown	Unknown	Unknown	Unknown	Unknown
Canary Islands, September 2002				
Unknown	Unknown	Unknown	Unknown	Unknown
Gulf of California, September 2002				
Air gun ³	Broadband	236-262dB re: 1µPa at 1m (p-p)	Vertical	No
Multi-beam sonar ⁴	15.5kHz	237dB re: 1µPa at 1m (RMS)	Omnidirectional	No
Sub-bottom profiler ³	3.5kHz	204dB re: 1µPa at 1m (RMS)	Vertical	No

Sources: ¹D'Amico and Verboom (1998); ²Anon. (2001); ³Federal Register (2003); ⁴Federal Register (2004). For more details, please see D'Spain *et al.* (2006).

and how this density may vary with season. However, we advocate a cautious approach when applying habitat models to regions that have not been thoroughly studied (i.e. extrapolating behaviour or habitat usage from one area to another) due, for example, to documented differences between known high density areas of beaked whales in different ocean basins (Barlow *et al.*, 2006; Ferguson *et al.*, 2006).

Anatomy and physiology

Rommel *et al.* (2006) reviewed the limited information available on anatomy and physiology of beaked whales. Given the scarce knowledge and the important conservation and mitigation implications, it is important that much more research be conducted on the anatomy and physiology of beaked whales, as well as the pathological changes caused by exposure to sound. We agree that emerging evidence supports the hypotheses that: (1) normal beaked whale diving patterns may lead to chronic tissue accumulation of nitrogen; and (2) chronic tissue accumulation of nitrogen may make beaked whales particularly vulnerable to diving related pathologies when their diving patterns are disrupted by exposure to intense sound. Research is needed in two key areas to further evaluate these hypotheses: (1) the factors contributing to nitrogen supersaturation, including normal and acoustically altered dive profiles and the depth at which complete lung collapse occurs; and (2) the potential for *in vivo* bubble nucleation and/or growth within tissues as a result of exposure to sound and/or disruption of normal diving patterns. In addition, the following are required: (1) better descriptions of normal gross and normal microscopic anatomies of healthy beaked whales (e.g. from incidental fishery takes and from 'normal' strandings); (2) investigations of the direct impacts of sound on tissues (*ex vivo*) presumed to be most susceptible to anthropogenic sound; (3) better descriptions of pathological changes in stranded beaked whales exposed to sound; (4) standardisation of gross and histopathological examination protocols for all beaked whale strandings, with special emphasis on the occurrence of gas and fat emboli and methods to prevent introduction of gas emboli during necropsies; (5) better descriptions of blood flow patterns in the vicinity of tissues potentially sensitive to sound; and (6) better descriptions of the anatomy and function of tissues and organs involved in hearing in beaked whales.

Comparative studies involving multiple beaked whale species and surrogate species (e.g. *Kogia*) may be useful. However, caution is required when extrapolating from other species to beaked whales. We therefore believe that when feasible, attempts should be made to rehabilitate live stranded beaked whales to provide opportunities for research not possible or more difficult with animals in the wild. However, it should be noted that an animal being held in rehabilitation will not experience the physiological challenges or adaptations associated with diving to depths of more than 500m; clearly observations made of a sick animal at the surface must be interpreted with caution.

Monitoring and mitigation

Barlow and Gisiner (2006) discussed the effectiveness of current monitoring and mitigation practices and described promising new tools for improving monitoring and mitigation in the near future. Current monitoring often involves a single observer using low-power (7×) binoculars searching for beaked whales and other marine mammals in all sea states during both day and night. Although it has been suggested that monitoring after dark may be aided with

recent night-vision technologies, this would require appropriate testing before being considered practical. Barlow and Gisiner (2006) provided a crude estimate that the visual methods currently employed may result in as little as a 1-2% chance of detecting beaked whales (the actual value will vary considerably depending on *inter alia* sea state and experience of observers). The present authors concur that these methods are ineffective in appreciably reducing interactions between beaked whales and potentially hazardous sound sources. Even using current best practices in visual surveys, such as those employed in line-transect abundance surveys with highly experienced observers, the probabilities of detecting beaked whales are 20-50% at best (Barlow and Gisiner, 2006). Passive acoustic sensors have not been used as part of beaked whale noise risk mitigation because little was known about their vocal behaviour. Recent data (Johnson *et al.*, 2004) indicate that passive acoustics may increase the probability of detecting beaked whales when the sensors are deployed at greater than 200m depth. However, any use of passive acoustic sensors at the surface must be tested carefully before being considered appropriate; it is possible beaked whales do not echolocate at shallower depths (Johnson *et al.*, 2004; Zimmer *et al.*, 2005). Other new sensing technologies such as active acoustics and radar have also not yet been tested sufficiently to assess their potential for detecting beaked whales.

Both long- and short-term research projects would help to better assess and mitigate the effects of anthropogenic sound on beaked whales. Important long-term studies include: (1) descriptions of population structure; (2) assessment of distribution and abundance for stocks and species; (3) development and testing of habitat use models; (4) assessment of population trends in local areas (e.g. in Abaco, Bahamas); and (5) systematic collection of information from live stranded and dead beaked whales. These studies would help to better identify sites of known or likely beaked whale occurrence, enable better assessment of the likely effects on individuals and populations from a given sound regime and lead to improved understanding of the clinical signs and pathologies of sound exposure. We also recommend the following short-term strategies: (1) detect and evaluate impacts of anthropogenic sound activities on beaked whales whenever a potential incident that may be a result of sound occurs; (2) conduct surveys for strandings and/or floating carcasses during and after anthropogenic sound activities; (3) determine the probability of detecting a floating carcass; (4) determine whether beaked whales avoid or approach vessels; and (5) incorporate behavioural reactions of beaked whales to anthropogenic sources of sound into monitoring measures.

POTENTIAL MECHANISMS

Although a number of beaked whale stranding events coincided with naval activities and active sonar use (e.g. Simmonds and Lopez-Jurado, 1991; Frantzis, 1998; Anon., 2001; Jepson *et al.*, 2003), the mechanism(s) by which sonar may lead to stranding and sometimes the death of beaked whales is not well understood. Determining such mechanisms is not only of scientific interest, but important in terms of mitigation. If, for example, the primary cause of strandings is a behavioural response in which whales avoid sound by moving into shallow water, then perhaps only those sound producing activities in close proximity to land need to be managed. Similarly, if these events resulted from abnormal acoustic propagation due to unusual

environmental conditions (e.g. waveguides – D'Spain *et al.*, 2006), then producers of sound need to monitor environmental conditions prior to introducing sound and mitigate when certain conditions occur. However, the available evidence is not currently sufficient to reach such conclusions.

Several possible mechanistic pathways through which sonar may lead to stranding and/or death of beaked whales are shown in Fig. 1. The first potential pathway entails a behavioural response to sound that leads directly to stranding, such as swimming away from a sound into shallow water. An alternative scenario involves a behavioural response leading to tissue damage. Such responses could include: a change in dive profile; staying at depth longer than normal; or remaining at the surface longer than normal. All of these responses could contribute to gas bubble formation, hypoxia, cardiac arrhythmia, hypertensive haemorrhage or other forms of trauma. Another pathway is through a physiological change such as a vestibular response leading to a behavioural change or stress induced hemorrhagic diathesis leading to tissue damage. Finally, beaked whales might also experience tissue damage directly from sound exposure, such as through acoustically mediated bubble formation and growth or acoustic resonance of tissues. Each of these potential mechanisms is described in detail below and at present it is not possible to rule out any of these.

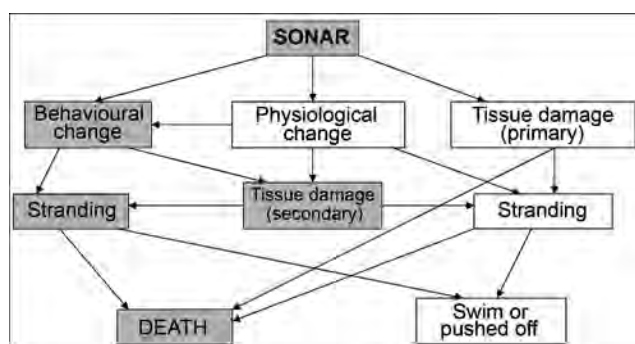


Fig. 1. Potential mechanistic pathways by which beaked whales are affected by sonar. Whereas we are unable to eliminate any pathways as implausible given current data, most of our discussions focus on the left side (shaded boxes) of the diagram. Note that death will not necessarily be the end result of sonar exposure in every case and that behavioural change, physiological change, primary tissue damage, secondary tissue damage, or stranding may occur without leading to death. This figure is intended to outline potential mechanisms of only those exposures which do lead to observed effect.

Behavioural response

Beaked whales may respond to sound by changing their behaviour, which could lead to a stranding event prior to the onset of physical trauma (Fig. 1, left). For example, in areas where deep waters occur in close proximity to shallow waters (e.g. 'canyon areas' of the Bahamas, oceanic islands), beaked whales may swim into shallow waters to avoid certain sounds and could strand if they are unable to navigate back to deeper waters. The end result of stranding may be that animals swim away, are pushed off, or die of hyperthermia or other stress-related causes resulting from the actual stranding. Evidence that some of the stranded beaked whales in the Bahamas succumbed to cardiovascular collapse due to hyperthermia (Anon., 2001) is consistent with this mechanism, although the final pathology report for this stranding event is still pending and the proposed

mechanism does not account for some of the trauma observed in that event (e.g. subarachnoid haemorrhage). The array of pathologies (Anon., 2001; Fernandez, 2004) observed in the beaked whales from the Bahamas and Canary Islands mass stranding in 2002 suggest injuries in addition to those typical of the physical effects of stranding itself.

Behavioural response leading to tissue damage

Acoustically induced behavioural responses may lead to tissue damage prior to stranding. Such responses may include altered dive profiles, remaining at the surface for prolonged periods or remaining at depth. Physiological responses could include hypoxia (from longer than normal time at depth or increased energy or oxygen use at a given time) or elevated nitrogen supersaturation of tissues, leading to formation of gas bubbles (from altered dive profiles).

One potential mechanism that deserves particular consideration is an acoustically induced behavioural change (dive response) that leads to formation of significant gas bubbles, which damage multiple organs or interfere with normal physiological function. Such a mechanism would be similar to decompression sickness in human divers and would have two parts: a dive response precipitating adverse gas bubble formation and pathology. Because many species of marine mammals make repetitive and prolonged dives to great depths, it has long been assumed that marine mammals have evolved physiological mechanisms to protect against the effects of rapid and repeated decompressions. To date, two physiological adaptations have been identified that may afford protection against nitrogen gas supersaturation: lung alveolar collapse at depths of 20-70m and 'elective circulation' involving vasoconstriction to the peripheral circulation during diving (Kooyman *et al.*, 1972; Ridgway and Howard, 1979; Zapol *et al.*, 1979; Davis *et al.*, 1983). However, Ridgway and Howard (1979), the only researchers who have assessed nitrogen gas accumulation in a diving cetacean, trained bottlenose dolphins (*Tursiops truncatus*) to dive repeatedly to 100m and found that the muscle of the dolphin was substantially supersaturated with nitrogen gas. From nitrogen washout curves, they estimated that this species experienced lung collapse at approximately 70m of depth, thus making it susceptible to nitrogen gas accumulation when making repetitive dives shallower than 70m. Houser *et al.* (2001) used the data from Ridgway and Howard (1979) to model the accumulation of nitrogen gas within the muscle tissue of other marine mammal species. The model was limited in that it necessarily assumed similar depths of lung collapse for all cetaceans and that exchange of nitrogen gas between tissue compartments ceased below the depth of lung collapse. The model predicted that those cetaceans that dive deep and have slow ascent/descent speeds would have tissues that are more supersaturated with nitrogen gas than other marine mammals. While the predictions for beaked whales were in excess of 300% supersaturation at the surface, this should be viewed cautiously because of the limitations of the model used and the problems of using extrapolations from other species.

Dive profiles of three species (Cuvier's and Blainville's beaked whales and bottlenosed whales, *Hyperoodon ampullatus*; Hooker and Baird, 1999; P. Tyack, unpub. data) suggest that at least some species of beaked whales have dive profiles not previously observed in other marine mammals. These led to the suggestion that some beaked whales may chronically accumulate nitrogen in a manner

not dissimilar to human ‘saturation divers’. The critical components of this dive sequence include: (1) very deep and long foraging dives (to as deep as 2km and lasting as long as 90mins); (2) relatively slow, controlled ascents, followed by (3) a series of ‘bounce’ dives to between 100–400m depth (Hooker and Baird, 1999; P. Tyack, unpub. data). Thus, if any part of this dive sequence was affected by a behavioural response to sound (e.g. extended time at surface without the requisite bounce dives), it could induce excessive levels of nitrogen supersaturation in tissues, driving gas bubble and emboli formation in a manner similar to decompression sickness in humans.

It is clear that long-term studies on the behaviour of beaked whales to better define a baseline of ‘normal’ behaviour are needed. Obtaining baseline dive profiles via several methods over extended periods (e.g. D-tags, time-depth recorders) is especially important. We unanimously agree that highest priority should be given to designing controlled-exposure experiments to investigate the responses of beaked whales to anthropogenic sounds. Nowacek *et al.* (2004) conducted a controlled exposure experiment in northern right whales (*Eubalaena glacialis*) responding to a novel alerting stimulus. This study demonstrated that whales responded to stimuli at received sound levels as low as 133dB (re: 1μPa), with an immediate ascent followed by an extended surfacing interval. It was hypothesised that abnormal changes in dive behaviour in beaked whales could precipitate pathologic bubble formation in tissues. By applying innovative technology, researchers can further investigate behavioural responses and begin to examine physiological responses to sound. Designing exposure studies that are acceptable from both a scientific and animal welfare perspective is difficult. We recommend that the best way to design such experiments is through a workshop of appropriate experts.

Determining whether beaked whales are susceptible to developing gas bubbles due to changes in behaviour or physiological condition may prove to be even more difficult. To date, while there is no evidence of *in vivo* bubble formation in any marine mammals (but see Jepson *et al.*, 2003; 2005; Fernandez *et al.*, 2004), it is also true that no studies have been conducted to specifically look for the formation of intravascular bubbles during or following repetitive diving. Although it is possible to conduct such studies with shallow diving species such as bottlenose dolphins, until such work is conducted with deep-diving species such as beaked whales, it will not be possible to gain an insight into this possibility. As noted above, marine mammals have long been thought to have evolved anatomical, physiological and possibly behavioural adaptations to their marine environment to mitigate the risk of bubble formation (e.g. Harrison and Tomlinson, 1956; Ridgway and Howard, 1979; 1982; Falke *et al.*, 1985; Ponganis *et al.*, 2003). Despite these adaptations, recent theoretical and pathological evidence suggests that cetaceans can produce *in vivo* bubbles or experience tissue injury as a result (Jepson *et al.*, 2003; 2005; Fernandez *et al.*, 2004; 2005). These data and interpretations are the subject of continuing scientific debate (Fernandez *et al.*, 2004; Piantadosi and Thalmann, 2004).

Modelling predictions (Houser *et al.*, 2001) support the hypothesis that beaked whale tissues could be greater than 300% saturated with nitrogen. *Post mortem* evidence of acute and chronic gas emboli-associated lesions in liver, kidney, spleen, and lymph nodes of eight dolphins, one harbour porpoise (*Phocoena phocoena*) and one Blainville’s beaked whale that stranded in the United Kingdom (Jepson

et al., 2003; 2005) also support this hypothesis. In addition, gas and fat emboli and widely disseminated microvascular haemorrhages were found in ten beaked whales examined in the Canary Islands mass strandings event in September 2002 (Jepson *et al.*, 2003; Fernandez *et al.*, 2004; 2005). In humans and experimental animals, such gas and fat emboli released into the venous system and deposited in the pulmonary capillary beds may travel through arterio-venous shunts into the systemic circulation. Prior to, or concomitant with this, respiratory and cardiovascular dysfunctions may occur with a biphasic response at the brain-spinal cord level, an initial, venous embolic obstruction and vasoconstriction, followed by secondary vasodilatation and prolonged (reactive) hyperaemia (Shigeno *et al.*, 1982). This haemodynamic process may explain the widespread cerebral congestion and edema, with spongiosis, intracranial perivascular haemorrhages and subarachnoid and intraventricular haemorrhages described in the beaked whale mass strandings.

A number of areas of research are required to further investigate whether beaked whales are susceptible to gas bubble formation, either as a function of altered behaviour or as the direct impact of sound on existing bubble nuclei. It is important that detailed necropsies are conducted of all freshly dead beaked whales, especially those whose deaths are correlated in both space and time with sound events. These necropsies should be conducted under laboratory procedures with rigorous protocols, e.g. opening the braincase underwater before the head is separated from the body or tying off primary vessels prior to removal, so as to avoid introducing bubbles during the necropsy. A standardised protocol for beaked whale necropsies is being developed to address these needs.

Experimental studies are needed to determine whether marine mammals can develop *in vivo* gas bubbles due to alterations in their dive profiles and to document precise levels of nitrogen supersaturation necessary to invoke such bubble formation. Specifically, ascertaining the onset of lung collapse and its impact on nitrogen gas kinetics is critical to determining what physiological effects any changes in dive profile might have on tissues. In the absence of live beaked whales for such studies, comparative studies could be conducted using marine mammals that are accessible and trainable. Physiological effects in shallow diving cetaceans and deep divers should be compared cautiously as deep divers will experience different physiological demands from environmental conditions at 1–2km and will likely have different adaptations and responses to those conditions. The depth at which lung collapse occurs is key to any modelling of nitrogen supersaturation because lung collapse prevents gas exchange and nitrogen absorption by the blood. The depth at which the lung collapses might be estimated using bottlenose dolphins with arterial blood sampling and blood nitrogen analyses. These results could then be compared to *post mortem* determination of lung collapse by compression testing of the lungs of a bottlenose dolphin carcass. Comparable results would support the use of *post mortem* testing of beaked whale lungs to determine the depth at which lung collapse occurs in those species. To test the hypothesised scenario that adverse gas bubble formation may result from a change in dive behaviour, it will be necessary to determine whether beaked whale tissues supersaturate with nitrogen and if so, to combine that information with dive profiles and potential changes in dive profiles. The scenario of gas bubble formation secondary to a behavioural response is plausible and merits rigorous investigation.

Physiological change

Haemorrhagic diathesis

The Bahamas beaked whale report listed twelve possible causes for the lesions observed and one proposed mechanism was haemorrhagic diathesis (Anon., 2001; 2002). Haemorrhagic diathesis is a tendency to bleed that results from one or more of several conditions, including: (1) depletion of clotting factors (disseminated intravascular coagulation (DIC)); (2) a hereditary deficiency in one or more of a suite of blood clotting factors; or (3) platelet dysfunction or thrombocytopenia. Humans with the hereditary deficiency develop haemorrhages in regions similar to those of the beaked whales (i.e. subarachnoid spaces and the inner ear; Palva *et al.*, 1979) and hypertension increases the likelihood of such patients suffering intracranial bleeds (Hart *et al.*, 1995). If beaked whales are subject to haemorrhagic diathesis, stress caused by exposure to sound may cause them to haemorrhage. Similar haemorrhages in human patients can cause headache, nausea and vomiting, confusion, ataxia, dizziness, loss of consciousness and even death (Hart *et al.*, 1995). By analogy, intracranial haemorrhages observed in beaked whales may have resulted in disorientation, a subsequent inability to navigate and eventual stranding (Anon., 2001).

While nothing is known currently about clotting abilities or DIC in beaked whales, a lack of clotting factors has been noted in some cetacean and pinniped species, which may be related to diving adaptations. Northern elephant seals (*Mirounga angustirostris*) have platelets that are less prone to triggering clotting at high pressure, such as at depth (Field *et al.*, 2001) and they are prone to DIC (Gulland *et al.*, 1996). Lack of certain clotting factors, specifically Hageman's factor, Fletcher factor activity and Factor IX, are common to all of the limited number of cetacean species studied to date (Lewis *et al.*, 1969; Robinson, A.J. *et al.*, 1969; Saito *et al.*, 1976). If all cetaceans lack multiple clotting factors, it is not clear why beaked whales exposed to sonar might be more susceptible to the effects of haemorrhage than other species. However, the fact that few other species stranded simultaneously in cases involving sonar may in part be a reflection of differences amongst species' perceptions of an event as stressful, fundamental susceptibilities to stress, or differences in subsequent responses to the event. Future studies are needed on the haematology and physiology of coagulation in beaked whales to determine whether they are predisposed to haemorrhaging. In addition, future studies should investigate differences in behavioural responses of beaked whales to stressful stimuli.

Vestibular response

Marine mammals could become disoriented due to a vestibular response to sounds. Tullio's phenomenon, or dizziness induced by sound, has long been known of in humans (Tullio, 1929). The peripheral vestibular system of beaked whales may be affected by sound, affecting their ability to navigate. Beaked whales, which are usually found in deep waters, might, if disoriented, move into shallow waters and be unable to navigate back to deeper waters. However, Balcomb and Claridge (2001) observed that when pushed towards deep water, several animals swam away without the characteristic rolling or turning movements typical of animals with vestibular pathology. Furthermore, disorientation can result from a number of phenomena, making it difficult to detect and attribute a vestibular response to sound exposure in the presence of other potentially contributing factors.

Primary tissue damage leading to behavioural response

Sound may damage tissue directly through acoustically mediated bubble growth or tissue shear. A scientific workshop organised by the US NOAA/National Marine Fisheries Service was held in 2002 to consider the potential for resonant effects of sound to induce tissue injury in cetaceans (Anon., 2002). Modelling of acoustic resonance in lungs of cetaceans and comparative data from other animal systems (e.g. humans, dogs, pigs) suggested that only minimal tissue injury is likely to result from such a mechanism because tissue displacements are minute (Anon., 2002). The only exception is the large excursions of tissue that could occur where two dramatically mismatched tissue boundaries intersect in which there was minimal damping by associated tissues. Discussions also occurred on the possibility of a mechanism of sonar-related tissue injury in cetaceans from acoustically mediated bubble growth, particularly in tissues supersaturated with nitrogen, as may occur towards the end of a dive (Anon., 2002). This concept was primarily based on the work of Crum and Mao (1996) and Houser *et al.* (2001). Crum and Mao (1996) modelled the likelihood of acoustically driven bubble growth in humans and marine mammals by the process of rectified diffusion. The model assumed modest levels of nitrogen tissue (super)saturation and predicted that relatively high sound pressure levels (>210dB re:1µPa) would be necessary to induce significant bubble formation in human divers or marine mammals at 300-500Hz. Houser *et al.* (2001) estimated that levels of nitrogen supersaturation in some tissues of some deep-diving species, such as the northern bottlenose whale, could exceed 300% near the surface, raising the possibility that acoustically mediated bubble formation might occur at received sound pressures and sound durations lower than those predicted by Crum and Mao (1996). The workshop therefore recommended that the Crum and Mao model (1996) be used to estimate the threshold sound pressure levels for the higher levels of nitrogen tissue supersaturation predicted to occur from typical beaked whale dive profiles (Anon., 2002).

Isolated porcine liver tissue, polyacrylamide gels and human blood that have been compressed 4-7 atmospheres for 1-3hrs and then decompressed to ambient show extensive bubble development when exposed to high intensity (230dB SPL re:1µPa) ultrasound of 37kHz (Crum *et al.*, 2005). The authors postulated that the underlying mechanism might be destabilisation of pre-existing bubble nuclei by the ultrasound exposure, resulting in bubble growth by static diffusion in supersaturated tissue. Although these experiments demonstrated a possible mechanism by which bubble growth might occur, it did so under conditions that are different from those to which beaked whales may have been exposed during the stranding events. Thus, it is premature to judge acoustically mediated bubble growth as a potential mechanism and we recommend further studies to investigate this possibility. Further exposure studies should be conducted on marine mammal tissues by saturating them, exposing them with frequencies and amplitudes of interest and testing for minimum levels that could result in tissue damage.

Acoustic resonance

Anon. (2002) also considered the possibility that beaked whales are susceptible to effects of acoustic resonance (see discussion above). Most participants agreed that the best available models indicated that acoustic resonance is highly unlikely in the lungs of beaked whales, but recommended further studies to fully eliminate this hypothesised

mechanism. They did not evaluate the possibility of resonance in other organs or structures and therefore recommended further modelling to determine if those would be susceptible to resonance. Given the full discussion in Anon. (2002), this mechanism is not discussed in depth here. The authors do, however, endorse the three areas of study recommended in Anon. (2002): (1) the possibility of resonance in the lung throughout the dive profile of beaked whales; (2) the potential for other organs or structures to be affected by acoustic resonance (either through modelling or empirical observation); and (3) the possibility that animals experience tissue shear (and determine how such injuries might appear).

Primary tissue damage leads to death

Some of the above mechanisms (i.e. gas bubble disease, haemorrhagic diathesis, acoustic resonance) could lead to lethal tissue damage. For example, the intracranial haemorrhage seen in the Bahamas and Canary Islands animals could have been caused by a stress response and associated haemorrhagic diathesis or bubble formation rupturing local capillaries. Although some of the stranded beaked whales were found dead, it is not clear whether these animals were alive when they first stranded. Several animals in all the events stranded alive and some either swam away or were pushed offshore. Even though their eventual fate is unknown, they did not die immediately. Determining whether sound exposure causes tissue damage that leads directly to death will be difficult and likely will require a process of elimination regarding other possible mechanisms. Testing the hypothesis that death results directly from sound-related tissue damage will be facilitated greatly by access to freshly stranded specimens that have been exposed to sound.

EDUCATION AND COORDINATION

As discussed below, education, communication and co-ordination will all facilitate the investigation of the effects of sound on beaked whales and mitigation measures to avoid adverse effects.

Education

Greater public outreach and education can be achieved through: (1) improved communication with environmental non-governmental organisations; (2) established links among scientists, the public and local and state policymakers; and (3) increased dissemination of stranding response information to the general public.

Co-ordination and communication

Improved co-ordination and communication is required among: (1) stranding responders to develop an international standardised protocol for necropsy; (2) sound producers, stranding responders and researchers to facilitate planning and preparation prior to sound exposure events and to monitor animal behaviour opportunistically; (3) sound producers and researchers to conduct retrospective analyses; (4) stranding responders to provide comprehensive databases to the public; (5) scientists and museums to obtain genetic samples from museum collections to evaluate population structure; and (6) terrestrial mammal and marine mammal physiologists to increase understanding of beaked whale physiology. Interaction across scientific disciplines (e.g. human dive physiology, terrestrial mammalogy, marine

mammal behaviour, etc.) is critical to an improved understanding of this problem and broad research co-ordination and co-operation are needed.

CONCLUSIONS

Monitoring and mitigation

Current visual survey efforts to detect beaked whales in areas of acoustic activity are probably ineffective as a mitigation aid. Key limiting factors include sea state, amount of daylight, experience of observers and the diving and surfacing behaviour of beaked whales, which makes them either difficult to see or unavailable for visual observation at the surface for long periods of time. For the same reasons, surveys to determine distribution and abundance are also difficult and limited in their reliability. However, additional sensing technologies, such as passive acoustics, active sonar and radar, are currently in development that may increase scientists' abilities to detect beaked whales. Improved baseline data on distribution, abundance and habitat preferences of beaked whales are needed, in addition to increased effort in detection and recovery of dead and injured animals for improved understanding of the effects of anthropogenic sound.

Research

Although no potential mechanisms can be eliminated at this stage, we highlight gas bubble formation mediated through a behavioural response as plausible and in need of intensive study. Intensive research is needed to eliminate or confirm this hypotheses. The following four research priorities will provide better insights into its possible role.

- (1) Controlled exposure experiments should be the top research priority. These experiments are critical for investigating beaked whale responses to sound. A multi-disciplinary workshop is needed to co-ordinate and design these experiments.
- (2) There is an urgent need for studies of anatomy, physiology and pathology of beaked whales, particularly in situations where there is a known cause of death (e.g. bycatch). A comprehensive, standardised necropsy protocol is needed to make the best possible use of animals that become available through stranding or fisheries interactions.
- (3) Baseline descriptions of diving behaviour and physiology of beaked whales are required to be able to better evaluate the potential for beaked whales to experience gas bubble disease from changes in dive behaviour.
- (4) Finally, a retrospective review of all stranding records is necessary, as well as new studies in areas beaked whales are concentrated and exposed to anthropogenic sounds. To the greatest extent possible, retrospective analyses should: (1) describe and compare pathologies from all stranding events; (2) model the received sound level at sites where sound-related stranding occurred; (3) document all anthropogenic sound sources during stranding events; (4) assess population level effects in areas where sufficient data are available (e.g. the Bahamas); (5) evaluate distribution of all strandings relative to surrounding oceanographic/topographic features and possibly-related anthropogenic sound activities; and (6) identify areas where beaked whales are present and naval exercises have occurred, but strandings have not been documented and compare those situations with documented stranding events.

Reviews should not interpret lack of strandings as sufficient evidence of no effect, because animals that die offshore may not wash ashore, animals that strand may not remain on the beach for more than one tidal cycle (Taylor *et al.*, 2004) and observation effort can vary markedly by location. Furthermore, whether or not strandings occur, activities involving anthropogenic sounds that may affect beaked whales should be documented to identify common features of habitat, species present or involved and acoustic properties to facilitate management and mitigation of such activities.

Understanding and evaluating potential mechanisms will aid managers in knowing when, where and how to best mitigate interactions between anthropogenic sound and beaked whales. The interdisciplinary approach of the workshop greatly facilitated exchanges of knowledge among scientists of disparate disciplines. The importance of interdisciplinary co-ordination and communication in solving this environmental problem cannot be overemphasised.

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